CLAIMS

1. A method of detecting and isolating cells that produce any secreted protein of interest (POI), comprising:

a) constructing a cell line transiently or stably expressing a cell surface capture molecule, which binds the POI, by transfecting the cell line with a nucleic acid that encodes such cell surface capture molecule;

b) transfecting said cell simultaneously or subsequently with a second nucleic acid that encodes a POI wherein such POI is secreted;

c) detecting the surface-displayed POI by contacting the cells with a detection molecule, which binds the POI;

d) isolating cells based on the detection molecule.

2. The method of claim 1 wherein the protein of interest is a ligand, a soluble receptor protein, or a growth factor.

3. The method of claim 1 wherein the protein of interest is an antibody, an Fab, a single chain antibody (ScFv), a fragment thereof, or anything fused to an antibody constant region.

4. The method of claim 2 wherein the growth factor is selected from the group consisting of Interleukin (IL)-1, IL-2, IL-4, IL-5, IL-6, IL-7, IL-9, IL-10, IL-13, IL-15, IL-16, IL-17, IL-18, IL-21, Ciliary Neurotrophic Factor (CNTF), erythropoietin, Vascular Endothelial

Growth Factor (VEGF), angiopoietin 1, angiopoietin 2, TNF, Interferon-gamma, GM-CSF, TGFβ, TNF Receptor, fusion proteins, and all approved therapies made in animal cells.

5. The method of claim 3 wherein the antibody is selected from the group consisting of IgM, IgG, IgA, IgD or IgE, as well as various subtypes of these.

6. The method of claim 1 wherein the nucleic acid that encodes a POI is selected from a DNA library.

7. The method of claim 1 wherein the cell surface capture molecule is a ligand-specific receptor, a receptor-specific ligand, an antibody-binding protein, an antibody, an ScFv, a fragment thereof, anything fused to a constant region of an antibody, and a peptide from a phage display or peptide library and derivatives that bind the POI.

8. The method of claim 7 wherein the cell surface capture molecule is Ang1, And2, VEGF, Tie1, Tie2, VEGFRI (Flt1), VEGFRII (Flk1), CNTF, CNTFR-alpha, cytokine receptor components, fusions of two or more cytokine receptor components, or any fragments thereof.

9. The method of claim 7 wherein the antibody binding protein is an Fc receptor, anti-immunoglobulin antibodies, anti-immunoglobulin ScFv, Protein A, Protein L, Protein G, Protein H or functional fragments thereof.

10. The method of claim 7 further comprising adding a membrane anchor to a protein such that it remains anchored in a cell

membrane, exposed to the outside of the cell, and functions as a cell surface capture molecule.

11. The method of claim 10 wherein the membrane anchor is a transmembrane anchor or a GPI link.

12. The method of claim 10 wherein the membrane anchor may be native to the cell, recombinant, or synthetic.

13. The method of claim 7 wherein a signal sequence is added to the amino terminus of a protein, such that the protein is transported to the cell surface, and functions as a cell surface capture molecule.

14. The method of claim 13 wherein the signal sequence may be native to the cell, recombinant, or synthetic.

15. The method of claim 1 wherein the isolated cell in claim 1(d) is an antibody producing cell fused to an immortalized cell.

16. The method of claim 15 wherein the antibody producing cell is a B-cell or derivative thereof.

17. The method of claim 16 wherein the B-cell derivative is a plasma cell, a hybridoma, a myeloma, or a recombinant cell.

18. The method of claim 1 wherein the detection molecule(s) are two molecules that bind each other and are differentially labeled.

19. The method of claim 1 wherein a blocking molecule which binds the cell surface capture molecule or POI is added to reduce the binding of the POI to a neighboring cell.

20. The method of claim 1 wherein the diffusion of the POI from the expressing cell to a neighboring cell and its adherence to that cell is reduced by increasing the viscosity of the media.

21. A non-human organism containing a cell produced by the method of claim 1.

22. A non-human organism containing a cell produced by the method of claim 1 wherein the cell surface capture molecule is specific for antibodies.

23. A method of detecting and isolating cells that produce any secreted protein of interest (POI), comprising:

a) constructing a cell line transiently or stably expressing a cell surface capture molecule, which binds the POI, by transfecting the cell line with a nucleic acid that encodes such cell surface capture molecule;

b) detecting a cell from (a) that expresses said cell surface capture molecule;

c) isolating and culturing the cell detected in (b);

d) transfecting said cell in (c) simultaneously or subsequently with a second nucleic acid that encodes a POI wherein such POI is secreted;

e) detecting the surface-displayed POI by contacting the cells with a detection molecule, which binds the POI;

f) isolating cells based on the detection molecule.

24. The method of claim 23 wherein the protein of interest is a ligand, a soluble receptor protein, or a growth factor.

25. The method of claim 23 wherein the protein of interest is an antibody, an ScFv, a fragment thereof, or anything fused to an antibody constant region.

26. The method of claim 24 wherein the growth factor is selected from the group consisting of Interleukin (IL)-1, IL-2, IL-4, IL-5, IL-6, IL-7, IL-9, IL-10, IL-13, IL-15, IL-16, IL-17, IL-18, IL-21, Ciliary Neurotrophic Factor (CNTF), erythropoietin, Vascular Endothelial Growth Factor (VEGF), angiopoietin 1, angiopoietin 2, TNF, Interferon-gamma, GM-CSF, TGFβ, TNF Receptor, fusion proteins, and all approved therapies made in animal cells.

27. The method of claim 25 wherein the antibody is selected from the group consisting of IgM, IgG, IgA, IgD or IgE, as well as various subtypes of these.

28. The method of claim 23 wherein the nucleic acid that encodes a POI is selected from a DNA library.

29. The method of claim 23 wherein the cell surface capture molecule is a ligand-specific receptor, a receptor-specific ligand, an antibody-binding protein, an antibody, an ScFv, a fragment thereof, anything fused to a constant region of an antibody, and a peptide from a phage display or peptide library and derivatives that bind the POI.

30. The method of claim 29 wherein the cell surface capture molecule is Tie1, Tie2, VEGFRI (Flt1), VEGFRII (Flk1), cytokine receptor components or fusions of two or more cytokine receptor components

31. The method of claim 29 wherein the antibody binding protein is an Fc receptors, anti-immunoglobulin antibodies, anti-immunoglobulin ScFv, Protein A, Protein L, Protein G, Protein H or functional fragments thereof.

32. The method of claim 29 further comprising adding a membrane anchor to a protein such that it remains anchored in a cell membrane, exposed to the outside of the cell, and functions as a cell surface capture molecule.

33. The method of claim 32 wherein the membrane anchor is a transmembrane anchor or a GPI link.

34. The method of claim 32 wherein the membrane anchor may be native to the cell, recombinant, or synthetic.

35. The method of claim 29 wherein a signal sequence is added to the amino terminus of a protein, such that the protein is transported to the cell surface, and functions as a cell surface capture molecule.

36. The method of claim 35 wherein the signal sequence may be native to the cell, recombinant, or synthetic.

37. The method of claim 23 wherein the isolated cell in (f) is an antibody producing cell fused to an immortalized cell.

38. The method of claim 37 wherein the antibody producing cell is a B-cell or derivative thereof.

39. The method of claim 38 wherein the B-cell derivative is a plasma cell, a hybridoma, a myeloma, or a recombinant cell.

40. The method of claim 23 wherein the detection molecule(s) are two molecules that bind each other and are differentially labeled.

41. The method of claim 23 wherein a blocking molecule which binds the cell surface capture molecule is added to reduce the diffusion of the POI from the expressing cell to a neighboring cell.

42. The method of claim 23 wherein the diffusion of the POI from the expressing cell to a neighboring cell and its adherence to that cell is reduced by increasing the viscosity of the media.

43. A non-human organism containing a cell produced by the method of claim 23.

44. A non-human organism containing a cell produced by the method of claim 23 wherein the cell surface capture molecule is specific for antibodies.

45. A method of detecting and isolating cells that produce a POI, comprising:

a) detecting a cell that expresses said cell surface capture molecule in high yield;

b) isolating and culturing the cell detected in (a);

c) transfecting said cell in (b) with a nucleic acid that encodes a POI wherein such POI is secreted;

d) detecting the surface-displayed POI by contacting the cells with a detection molecule which binds the POI;

e) isolating cells based on the detection molecule.

46. The method of claim 45 wherein the protein of interest is a ligand, a soluble receptor protein, a growth factor, or an antibody.

47. The method of claim 46 wherein the growth factor is selected from the group consisting of Interleukin (IL)-1, IL-2, IL-4, IL-5, IL-6, IL-7, IL-9, IL-10, IL-13, IL-15, IL-16, IL-17, IL-18, IL-21, Ciliary Neurotrophic Factor (CNTF), erythropoietin, Vascular Endothelial Growth Factor (VEGF), angiopoietin 1, angiopoietin 2, TNF, Interferon-gamma, GM-CSF, and TGFβ.

48. The method of claim 46 wherein the antibody is selected from the group consisting of IgM, IgG, IgA, IgD or IgE, as well as various subtypes of these.

49. The method of claim 45 wherein the nucleic acid that encodes a POI is selected from a DNA library.

50. The method of claim 45 wherein the cell surface capture molecule is a ligand-specific receptor, a receptor-specific ligand, or an antibody binding protein.

51. The method of claim 50 wherein the cell surface capture molecule is Tie1, Tie2, VEGFRI (Flt1), VEGFRII (Flk1), cytokine receptor components or fusions of two or more cytokine receptor components.

52. The method of claim 50 wherein the antibody binding protein is an Fc receptors, anti-immunoglobulin antibodies, anti-immunoglobulin ScFv, Protein A, Protein L, Protein G, Protein H, or functional fragments thereof.

53. The method of claim 45 further comprising adding a membrane anchor to a protein such that it remains anchored in a cell membrane, exposed to the outside of the cell, and functions as a cell surface capture molecule.

54. The method of claim 53 wherein the membrane anchor is a transmembrane anchor or a GPI link.

55. The method of claim 53 wherein the membrane anchor may be native to the cell, recombinant, or synthetic.

56. The method of claim 50 wherein a signal sequence is added to the amino terminus of a protein, such that the protein is transported to the cell surface, and functions as a cell surface capture molecule.

57. The method of claim 56 wherein the signal sequence may be native to the cell, recombinant, or synthetic.

58. The method of claim 45 wherein the isolated cell in (f) is an antibody producing cell fused to an immortalized cell.

59. The method of claim 58 wherein the antibody producing cell is a B-cell or derivative thereof.

60. The method of claim 59 wherein the B-cell derivative is a plasma cell, a hybridoma, a myeloma, or a recombinant cell.

61. The method of claim 45 wherein the detection molecule(s) are two molecules that bind each other and are differentially labeled.

62. The method of claim 45 wherein a blocking molecule which binds the cell surface capture molecule is added to reduce the diffusion of the POI from the expressing cell to a neighboring cell.

63. The method of claim 45 wherein the diffusion of the POI from the expressing cell to a neighboring cell and its adherence to that cell is reduced by increasing the viscosity of the media.

64. A non-human organism containing a cell produced by the method of claim 45.

65. A non-human organism containing a cell produced by the method of claim 45 wherein the cell surface capture molecule is specific for antibodies.

66. A method of detecting and isolating cells that produce high levels of POI, comprising:

a) constructing a cell line expressing a cell surface capture molecule, which binds the POI, by transfecting the cell line with a nucleic acid that encodes such cell surface capture molecule;

b) detecting a cell from (a) that expresses said cell surface capture molecule in high yield;

c) isolating and culturing those cells detected in (b) and allowing sufficient time for said cell to secrete the POI;

d) detecting the surface-displayed POI by contacting the cells with (a) detection molecule(s), one or more of which binds the POI;

e) isolating cells based on the detection molecule(s).

67. The method of claim 66 wherein the protein of interest is a ligand, a soluble receptor protein, a growth factor, or an antibody.

68. The method of claim 67 wherein the growth factor is selected from the group consisting of Interleukin (IL)-1, IL-2, IL-4, IL-5, IL-6, IL-7, IL-9, IL-10, IL-13, IL-15, IL-16, IL-17, IL-18, IL-21, Ciliary Neurotrophic Factor (CNTF), erythropoietin, Vascular Endothelial Growth Factor (VEGF), angiopoietin 1, angiopoietin 2, TNF, Interferon-gamma, GM-CSF, and TGFβ.

69. The method of claim 67 wherein the antibody is selected from the group consisting of IgM, IgG, IgA, IgD or IgE, as well as various subtypes of these.

70. The method of claim 66 wherein the nucleic acid that encodes a POI is selected from a DNA library.

71. The method of claim 66 wherein the cell surface capture molecule is a ligand-specific receptor, a receptor-specific ligand, or an antibody binding protein.

72. The method of claim 71 wherein the cell surface capture molecule is Tie1, Tie2, VEGFRI (Flt1), VEGFRII (Flk1), cytokine receptor components or fusions of two or more cytokine receptor components

73. The method of claim 71 wherein the antibody binding protein is an Fc receptors, anti-immunoglobulin antibodies, anti-immunoglobulin ScFv, Protein A, Protein L, Protein G, Protein H, or functional fragments thereof.

74. The method of claim 71 further comprising adding a membrane anchor to a protein such that it remains anchored in a cell membrane, exposed to the outside of the cell, and functions as a cell surface capture molecule.

75. The method of claim 74 wherein the membrane anchor is a transmembrane anchor or a GPI link.

76. The method of claim 74 wherein the membrane anchor may be native to the cell, recombinant, or synthetic.

77. The method of claim 71 wherein a signal sequence is added to the amino terminus of a protein, such that the protein is transported to the cell surface, and functions as a cell surface capture molecule.

78. The method of claim 77 wherein the signal sequence may be native to the cell, recombinant, or synthetic.

79. The method of claim 66 wherein the isolated cell in (f) is an antibody producing cell fused to an immortalized cell.

80. The method of claim 79 wherein the antibody producing cell is a B-cell or derivative thereof.

81. The method of claim 66 wherein the B-cell derivative is a plasma cell, a hybridoma, a myeloma, or a recombinant cell.

82. The method of claim 66 wherein the detection molecule(s) are two molecules that bind each other and are differentially labeled.

83. The method of claim 66 wherein a blocking molecule which binds the cell surface capture molecule is added to reduce the diffusion of the POI from the expressing cell to a neighboring cell.

84. The method of claim 66 wherein the diffusion of the POI from the expressing cell to a neighboring cell and its adherence to that cell is reduced by increasing the viscosity of the media.

85. A non-human organism containing a cell produced by the method of claim 66.

86. A non-human organism containing a cell produced by the method of claim 66 wherein the cell surface capture molecule is specific for antibodies.